

Claims

1. Recombinant seven-transmembrane receptor, whereby the amino terminus of said recombinant receptor is located on an extracellular side and the carboxy-terminus is located on an intracellular side of a membrane, comprising at least two detectable labels, whereby
 - a first of said at least two detectable labels is or is located on the carboxy-terminus and whereby a second of said at least two labels is or is located on the first or third intracellular loop; or whereby
 - a first of said at least two labels is or is located on the first intracellular loop and a second of at said at least two labels is or is located on the third intracellular loop.
2. Recombinant membrane receptor of claim 1, whereby said first label is or is located on the third intracellular loop of said membrane receptor and wherein said second label is or is located on the carboxy terminus.
3. Recombinant membrane receptor of claim 1 or 2, whereby said membrane receptor is a G-protein-coupled receptor or a proto-oncogene.
4. Recombinant membrane receptor of claim 1, whereby the proto-oncogene is Smoothed receptor (Smo) or whereby said G-protein-coupled receptor (GPCR) is selected from the group consisting of a rhodopsin/ β 2 adrenergic receptor-like GPCR, a glucagon/VIP/calcitonin receptor-like GPCR and a metabotropic neurotransmitter/calcium receptor.
5. Recombinant membrane receptor of claim 4, whereby said rhodopsin/ β 2-adrenergic receptor-like GPCR is the α 2A adrenergic receptor or the adenosine receptor A2A or wherein said glucagon/VIP/calcitonin receptor-like GPCR is the parathyroid hormone (PTH) receptor.

6. Recombinant membrane receptor of any one of claims 3 to 5, whereby said G-protein-coupled receptor or said proto-oncogene is of human or of mouse origin.
7. Recombinant membrane receptor of any one of claims 1 to 6, whereby said detectable labels are fluorescent labels or bioluminescent labels.
8. Recombinant membrane receptor of claim 7, whereby said fluorescence labels are selected from the group consisting of GFP, YFP, CFP, BFP, citrine, sapphire and dsRed, whereby said bioluminescent labels is luciferase (like renilla luciferase or firefly luciferase), or whereby said fluorescence label is produced by binding the FIAsh compound to specific epitopes of said 1st and 3rd loop or said C-terminus of the recombinant seven-transmembrane receptor.
9. Recombinant membrane receptor of any one of claims 3 to 8, whereby said G-protein-coupled receptor comprising at least two labels is selected from the group consisting of:
 - (a) a polypeptide as shown in SEQ ID NOS: 12, 14, 16, 40 or 42;
 - (b) a polypeptide encoded by a nucleic acid sequence as depicted in any one of SEQ ID NOS: 11, 13, 15, 39 or 41;
 - (c) a recombinant membrane receptor of any one of claims 3 to 8 encoded by a nucleotide sequence which hybridizes to a nucleotide sequence as defined (b); and
 - (d) a recombinant membrane receptor of any one of claims 3 to 8 encoded by a nucleic acid sequence being degenerate as a result of the genetic code to a nucleic acid sequence as defined in (b) or (c).
10. Recombinant membrane receptor of any one of claims 3 to 9, wherein the third intracellular loop being or comprising said first label is selected from the group consisting of
 - (a) a polypeptide depicted in SEQ ID NOS: 18, 22 or 26;

- (b) a polypeptide encoded by a nucleic acid sequence as depicted in SEQ ID NOS: 17, 21 or 25;
 - (c) a third intracellular loop encoded by a nucleotide sequence which hybridizes to a nucleotide sequence as defined (b); and
 - (d) a third intracellular loop encoded by a nucleic acid sequence being degenerate as a result of the genetic code to a nucleic acid sequence as defined in (b) or (c).
11. A nucleic acid molecule encoding the recombinant seven-transmembrane receptor of any one of claims 1 to 10.
12. A vector comprising a nucleic acid molecule as defined in any one of claims 8 to 11.
13. The vector of claim 12, which is an expression vector.
14. A host transformed with a vector of claim 12 or 13 or transfected with a nucleic acid molecule as defined in any one of claims 8 to 11.
15. The host of claim 14 which is a mammalian cell, an amphibian cell, a fish cell, an insect cell, a fungal cell, a plant cell or a bacterial cell, or a transgenic non-human animal.
16. The host of claim 15, wherein said mammalian cell is a CHO-cell or a HEK293 cell, a PC12 cell, a (primary) cardiomyocyte or a cultured neuronal cell.
17. The host of claim 15, wherein said amphibian cell is an oocyte, preferably a xenopus oocyte.

18. A method for producing a recombinant membrane receptor as defined in any one of claims 1 to 10 comprising culturing/raising the host of any one of claims 14 to 17 and, optionally, isolating the produced polypeptide.
19. A method for identifying molecules or compounds which are capable of activating, deactivating or inactivating the (biological/pharmacological) function of a seven-transmembrane receptor, comprising the steps of
 - (a) contacting the recombinant membrane receptor as defined in any one of claims 1 to 10 or a host or a host cell of any one of claims 14 to 17 with (a) molecule(s) or compound(s) to be tested; and
 - (b) measuring whether said molecule(s) or compound(s) to be tested lead(s) to a modification of a signal provided by said at least two detectable labels.
20. A method of screening for molecules or compounds which are activators (agonists) or inhibitors (antagonists) of the (biological/pharmacological) function of a seven-transmembrane receptor comprising the steps of
 - (a) contacting a recombinant membrane receptor as defined in any one of claims 1 to 10 or a host or a host cell of any one of claims 14 to 17 with the molecule or compound to be tested;
 - (b) measuring and/or detecting a response comprising a modification of a signal provided by said at least two detectable labels; and
 - (c1) comparing said response to a standard response as measured in the absence of said candidate molecule/compound
 - (c2) comparing said response to the response of a control membrane receptor which comprises at least two detectable labels on the C-terminus; or
 - (c3) comparing said response to control seven-transmembrane receptor which comprises only one detectable label.

21. A method for identifying molecules or compounds which are capable of eliciting a (biological/pharmacological) response of a seven-transmembrane protein, comprising the steps of
- (a) contacting a membrane protein of any one of claims 1 to 10, or a host or host cell of claims 14 to 17 with the molecule or compound to be tested; and
 - (b) identifying among these molecules/compounds the molecules/compounds which are capable of eliciting a change in energy emitted by said at least two detectable labels comprised on the recombinant membrane receptor as defined in any one of claims 1 to 10 .
22. The method of any one of claims 19 to 21, whereby said response or said energy change is an increase or a decrease of fluorescence resonance energy transfer (FRET).
23. The method of any one of claims 19 to 21, whereby said response or said energy change is an increase or a decrease of bioluminescent resonance energy transfer (BRET).
24. A diagnostic composition comprising the recombinant membrane protein of any one of claims 1 to 10 or the nucleic acid molecule as defined in claims 8 to 11, the vector of claims 12 or 13, the host cell of claims 14 to 17 or organs or cells of the non-human transgenic animal of claim 15.
25. A kit comprising the recombinant membrane protein of any one of claims 1 to 10 or the nucleic acid molecule as defined in claims 8 to 11, the vector of claims 12 or 13, the host cell of claims 14 to 17 or organs or cells of the non-human transgenic animal of claim 15.
26. Use of the recombinant membrane protein of any one of claims 1 to 10 or the nucleic acid molecule as defined in claims 9 or 10, the vector of claims

11 or 12, the host cell of claims 13 to 16 or organs or cells of the non-human transgenic animal of claim 16 for the detection of (a) modifier(s) of the biological activity of seven-transmembrane receptors in vivo or in vitro.